

THE HYPOPHYSIS OF THE BROAD-BILLED SWORDFISH, XIPHIAS GLADIUS L.

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The life history of the broad-billed swordfish (*Xiphias gladius L.*) is unknown, despite the investigations of marine biologists for a considerable period (Goode, 1873; Fish, 1926). Information concerning the habits of the adult fish, the location of its breeding grounds, or the occurrence of sexually mature swordfish is completely lacking. Therefore, a knowledge of the reproductive condition of the swordfish in all parts of its extensive range would be helpful in locating its breeding areas. The purpose of this paper is to determine primarily the aspect of the hypophysis of those swordfish which occur in southern New England waters. Their state of ovarian development has been described (Lee, 1942). It is hoped that these studies which have established the reproductive condition of those swordfish which are taken in this region, will be extended to include fish from all parts of its range, eventually locating its breeding grounds by this means.

The study of the hypophysis of the swordfish captured off the southern New England coastline was begun in the summer of 1940. Twenty-three pituitaries were obtained during this season, but the sex of the fish from which they were taken was undetermined. Therefore, the work was repeated in the summer of 1941 on glands from 13 swordfish, all of which proved to be female. Their pituitary glands were identical with those obtained during 1940.

METHODS

Dawson's and Friedgood's (1938) method of preparing the pituitaries of the rabbit and cat for study was used throughout this examination of the swordfish gland. This method was used successfully by Scruggs (1939) in his studies of the hypophyses of 16 different fish. The glands were fixed for 48 hours in ten parts of 0.1 per cent NaCl solution supersaturated with $HgCl_2$, to which one part of formalin was added just prior to use. Serial sections were cut at three to four micra.¹

* Contribution No. 317.

Owing to the conditions of obtaining the pituitaries, an interval of from four to six hours often occurred between the capture of the fish and the fixation of the gland. However, all classes of cells appeared maintained, cytoplasmic granules were preserved, the vacuolar membranes were intact and negative images of the Golgi apparatus were readily found. The fixation compared favorably with that obtained for other fish pituitaries fixed immediately by the same method (Scruggs, 1939) and with one swordfish pituitary fixed less than 40 minutes after capture of the fish. The relatively good preservation of the material was probably due to the fact that the heads were removed and packed in ice below decks upon capture of the swordfish.

RESULTS

The hypophysis lies beneath the infundibular region of the brain in a deep sella turcica, which is lined with periosteum and with that portion of the meninx that descends to form a tightly fitting sheath about the gland. As in higher organisms, the dorsal surface of the gland is covered by a diaphragm of the meninx, which is reflected to form a collar of connective tissue between the hypothalamus and the pituitary. There is no saccus vasculosus, nor any structure which may be likened to it, in the swordfish.

The pituitary gland is relatively large with respect to the brain. In four specimens chosen at random, the gland averaged 0.13 cc. in volume and weighed 0.08 gm., wet weight. In shape it resembles a blunt tetrahedron with its base pressed closely against the infundibulum. One surface faces anteriorly and the other two extend postero-medially to form the posterior margin of the gland. From the corners and apex of the hypophysis, extensions of the meninx adhere to or fuse with the periosteum, binding the gland tightly into the sella.

Sagittal and frontal sections of the pituitary reveal that the gland is divided into the four usual regions described by Stendell (1914) and other investigators of the teleost pituitary gland: the pars nervosa, the pars intermedia, the pars anterior, and the Übergangsteil; but the topographical distribution of these regions varies from the customary arrangement.

¹ The brain of the broad-billed swordfish is exceptionally small in relation to the size of the body. The average brain weight was 1.9 gm. and the average length was 3.5 cm. in four specimens chosen at random, whose average weight was 160 kg. and whose average total length was 2.8 m.

The pars nervosa

There is no discrete stalk of nervous tissue connecting the hypophysis to the brain. A slender tongue of the infundibular recess descends through the ventral medial sulcus, then expands laterally to form a small secondary cavity of an inverted mushroom shape. From the thin ventral wall of this cavity, which lies directly in contact with the dorsal surface of the hypophysis, the pars nervosa penetrates the pars glandularis. The pars nervosa appears primarily as a thick stalk of nervous tissue extending into the center of the hypophysis. Here it arborizes completely into a tree-like structure which is entirely surrounded by the pars intermedia. Accessory penetrations of tissue from the infundibular region project into the gland, peripherally to the main nervous stalk. The exact nature of these secondary penetrations is unknown, but they appear to be combinations of connective tissue and blood vessels from the meningeal diaphragm, with slender trabeculae of nervous tissue which descend directly from the infundibulum. The pars nervosa possesses the fibrous nature reported by Bock (1928), Charipper (1936), and others for this region of the teleost pituitary gland. The nuclei of connective tissue cells are common; but the chromatic granules, or Herring bodies, such as were found by Scruggs (1942) in the pars nervosa of the goldfish and carp, were not found in this portion of the swordfish pituitary.

The cells in the pars anterior of the mammalian pituitary are described as "acidophiles," "basophiles," or "chromophobes," depending upon whether they assume the so-called "acid" stains such as eosin, orange G, or azocarmine, the so-called "basic" stains such as methyl blue, or assume no stain whatever. This terminology has been transferred to cells within the three subdivisions of the teleost pituitary gland, which react in apparently similar fashion, by Matthews (1936), Scruggs (1939), Kerr (1940) and other investigators. A like procedure has been adopted in the description of the component cells of the swordfish hypophysis. Those pars intermedia cells which stain with orange G are referred to as "pars intermedia acidophiles," etc.

The pars intermedia

The pars intermedia appears as a cone-like region with its apex directed ventrally which completely surrounds the arborized portion of the main nervous penetration. This region of the hypophysis is characterized by predominant weak basophiles which assume a dull blue color. These cells are small, being six to eight micra in diameter, and

are generally iso-diametric. Occasionally basophiles are found in the pars intermedia which are twice the usual size and possess small carminophilic granules distributed throughout the cytoplasm. These granules may be mitochondria, for those in proximity to the nucleus often assume rod-like forms. In addition to the basophiles, orange G acidophiles are also found in the pars intermedia, but these cells are much fewer in number. The cytoplasm of the acidophiles is of a dull orange color, with a fine granulation. Negative images of the Golgi apparatus are often seen in this type of cell. There is no definite arrangement of the acidophiles within the pars intermedia of the swordfish, as Scruggs (1939) and Bell (1937) found in the goldfish hypophysis, for they seem scattered at random throughout this part of the gland.

Degranulated cells, with an indistinct outline and a partially pycnotic nucleus, occur throughout the pars intermedia. These strongly resemble similar cells described by Scruggs (1939) for the pars intermedia of the majority of teleost pituitaries that he studied.

The Übergangsteil, or "transitional region"

The main portion of this part of the hypophysis is wedge-like in sagittal section, and lies dorsally to the base of the cone-shaped pars intermedia, with the main mass of tissue in the postero-dorsal corner of

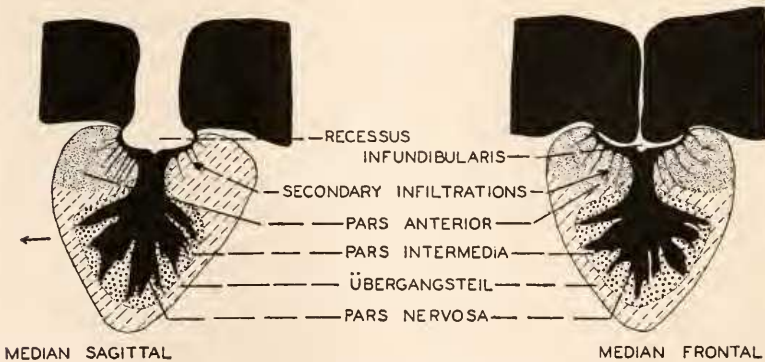


FIG. 1. Diagrams of median sagittal and median frontal sections of the swordfish pituitary gland, showing the topographical arrangement of the glandular regions.

the gland (Fig. 1). From this region paired processes extend anteriorly around the main nervous penetration to the frontal surface of the pituitary. A sheath of Übergangsteil material also descends over the pars intermedia, completely enveloping it and forming the superficial layer

of the hypophysis, excepting that portion of the gland where the pars anterior is situated.

The cells of the Übergangsteil are of two types; one is an acidophile and the other a basophile. The acidophiles are strong carminophiles, which react readily with and retain the azocarmine to assume a deep crimson color. Their cytoplasm is finely granulated, with larger granules dispersed through the cell. Usually the acidophiles are clustered about the capillaries, which are most numerous in this region of the gland; they also border the penetrations of infundibular material. The basophiles of the Übergangsteil, in those fish captured in midsummer, are of a brilliant blue color, with homogeneous cytoplasm and an exceptionally large cytoplasmic vacuole in each cell. (See plate.) The entire Übergangsteil appears net-like in sagittal section, due to the presence of these vacuolated basophiles.

The pars anterior

The pars anterior occupies a horse-shoe shaped region in the anterodorsal portion of the hypophysis, with its arms extending posteriorly around the zone of contact of the infundibulum with the pars glandularis to meet the main Übergangsteil mass. There are two types of cells, an amphiphile and an acidophile, in the pars anterior of the swordfish. The amphiphiles react polychromatically with the triple stain, showing a predominant tendency to assume the anilin blue which gives them a bluish-purple color. These cells line the infiltrations of infundibular material which penetrate this part of the gland. Their nuclei adopt a distal position with regard to the infundibular tissue; the intervening cytoplasm is very finely granulated and usually possesses small vacuoles.

The bulk of the pars anterior is composed of dark red carmine acidophiles. These are readily distinguished from the type of acidophiles in the Übergangsteil by means of their predominant color and coarsely granulated cytoplasm. In those acidophiles which approximate the numerous capillaries in the pars anterior, there is a definite concentration of these granules in the ends of the cells proximal to the blood vessels, with the nuclei occupying a distal position.

Histological Changes in the Pars Glandularis

During both summers that the swordfish pituitaries were studied, considerable changes were found in the aspect of the pars intermedia and the Übergangsteil, as the season progressed.

The pars intermedia of pituitaries obtained in early July possessed one to two orange G acidophiles per high power microscopic field ($490\times$). The numbers of these cells gradually increased in pituitaries obtained successively throughout the summer, until there were 10 to 11 such acidophiles present per high power field in the pars intermedia of the last fish captured in early September. As the acidophiles increased in number, they appeared to be most numerous in the regions of the pars intermedia closest to the main branches of the pars nervosa which it surrounds. In this respect, the pars intermedia and its components in the swordfish agree more closely with the findings of Bell (1937) and Scruggs (1939) mentioned previously.

The basophiles of the Übergangsteil also showed marked changes in appearance during both summers. The aspect of these cells in pituitaries taken from late July to the end of the season has been described previously. However, there is a relatively complete absence of vacuoles in the Übergangsteil basophiles of pituitaries obtained in early July, and the cytoplasm of these cells possesses a coarse granulation. (See Plate.) As the sex of those swordfish from which these pituitaries were taken was undetermined, the exact nature and causes of the differences in aspect of the Übergangsteil basophiles still remain to be investigated.

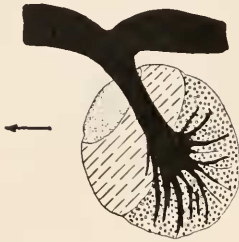
DISCUSSION

The topographical appearance of the swordfish hypophysis presents an interesting comparison with that of teleost pituitary glands that have been studied previously (Stendell, 1914; Bock, 1928; Charipper, 1936; Bell, 1937; Scruggs, 1939) with regard to the Übergangsteil. The generalized teleost pituitary is composed of two primary regions: that of nervous origin, the pars nervosa, and that of buccal origin, the pars glandularis. In the latter, the Übergangsteil develops as a plate or disc-like collar around the more dorsal portion of the pars nervosa, situated more or less completely between the pars intermedia and the pars anterior. However, in the swordfish the Übergangsteil is considerably flattened in its frontal portion, with the bulk of its tissue in the postero-dorsal region of the gland. A sheath of its components also invests completely the pars intermedia, to form the superficial layer of the hypophysis (Fig. 2). This condition suggests that in organogeny the process of intergrowth and differentiation of the pars glandularis with the pars nervosa is incomplete, as compared to the same process in other teleosts. Scruggs (1939) found a topographical condition similar to that of the swordfish hypophysis in the *Centrarchidae*; but in these fish the Über-

gangsteil sheath is fragmentary or completely lacking in the postero-medial line.

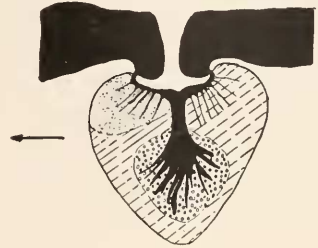
Because of differences in methodology, it is extremely difficult to compare the cellular components of the swordfish hypophysis with those of other teleosts that have been studied. A comparison of the nature of the subdivisions in the pars glandularis is probably valid, however, if made with regard to the predominant cell types present (basophiles, chromophobes, etc.), and to their distribution within the region in which they occur. Scruggs (1939) used the azan technique extensively on teleost pituitaries; hence the cellular components of the swordfish pitui-

GENERALIZED HYPOPHYSIS
OF TELEOST



P. NERVOSA —■—
P. INTERMEDIA —▨—

SWORDFISH
HYPOPHYSIS



P. ANTERIOR —▨—
ÜBERGANGSTEIL —▤—

FIG. 2. Diagrams comparing the topographical arrangement of the glandular regions in the swordfish pituitary gland with that in the generalized teleost pituitary. (Anterior end to the left.)

tary gland may be compared more closely with those of the teleost glands which he studied.

The cell types of the pars intermedia compare favorably with those described by Charipper (1936), Bell (1937) and Scruggs (1939). Weak basophiles characterize the region, with weak acidophiles scattered through them, more or less at random. Occasional chromophobes are evidently formed by degranulation of the basophilic cells. The fact that Dawson (1940) found these three types of cells in the pars intermedia of *Protopterus aethiopicus* indicates that this region of the fish pituitary gland is relatively constant with regard to its cell components.

The components of the Übergangsteil also agree with the general condition found by other students of the teleost pituitary gland. Its brilliant carminophiles and deep blue basophiles are essentially the same

types of cells that Scruggs (1939) found in the Übergangsteil of all fish that he studied save the flounder and stickle-back. Charipper (1936), Kerr (1940) and others are agreed that this part of the teleost hypophysis is usually highly chromatic, possessing strong acidophiles and basophiles with the various staining techniques.

The predominantly chromatic nature of the pars anterior, however, is at variance with the condition that Scruggs (1939) describes for the large majority of fish that he examined. Bell (1937) has shown that the pars anterior of the goldfish is dominated by orange G acidophiles; while Kerr (1940) found this region of the trout pituitary to be chiefly acidophilic, possessing a brownish-red acidophile when stained with Mallory's triple stain. Thus the pars anterior of the teleost pituitary gland may vary considerably in different species and genera.

In its fibrous appearance the pars nervosa of the swordfish hypophysis resembles that of other fish that have been examined; however, the chromatic granules described by Scruggs (1939, 1942) for the pars nervosa of many species, and by Evans (1940) for the eel, are not found in the swordfish.

The changes in aspect of the Übergangsteil and pars intermedia of the swordfish hypophysis are without precedence in the literature. However, several investigators have reported various dissimilar phenomena in the pituitaries of other fish. Matthews (1939) and Scruggs (1939) have found several changes in the pituitary gland of *Fundulus heteroclitus* that are correlated with the beginning of the breeding season. Scruggs (1942) describes marked changes in the appearance of the pituitaries of *Cyprina carpio* and *Carassius auratus* that appear prior to and during the breeding periods of these fish. Evans (1940) reports large colloid-filled sinusoids in the pars intermedia of the eel, appearing

PLATE I

FIG. 3. The forward process of the Übergangsteil of a swordfish pituitary gland obtained on July 12, 1941. The darker cells are the bright crimson carmine acidophiles; the lighter cells are the granular basophiles. (720 X.)

FIG. 4. The forward process of the Übergangsteil of a swordfish pituitary gland obtained on August 4, 1941. The basophiles possess large cytoplasmic vacuoles. Their nuclei are slightly pycnotic, and their cytoplasm has lost its granulation, to assume a dense homogeneous nature. This condition prevails for the remainder of the season. (800 X.)

FIG. 5. The layer of amphiphiles in the pars anterior of a swordfish captured on July 24, 1940. The amphiphiles border an infiltration of connective tissue and blood vessels from the infundibular region. (1080 X.)

FIG. 6. Carmine acidophiles bordering a blood vessel in the pars anterior of a swordfish captured on July 24, 1940. (The same fish whose pars anterior is shown in Figure 5.) The large carminophilic granules are concentrated in that end of the cells nearest to the blood vessel. (1080 X.)

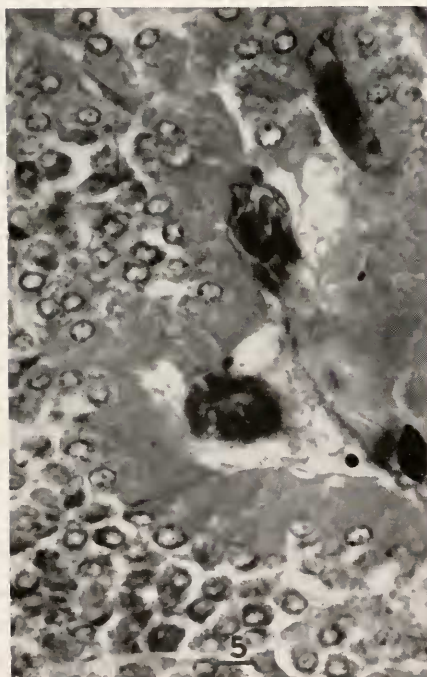
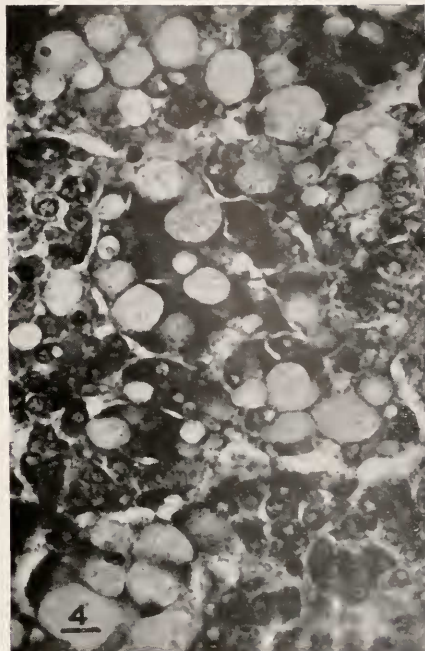
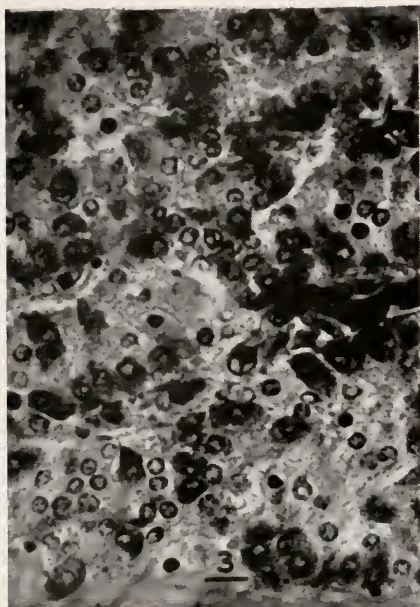


PLATE I

just before the onset of the migration impulse, with an enlargement of the pars anterior found at approximately the same time. Although these changes are not similar to those found in the pituitary of the swordfish, they indicate the components of the teleost pituitary gland may undergo a considerable modification which is apparently correlated with the breeding period. As the ovaries of the swordfish show no indication of maturation or of an increased pituitary activity during the brief time that the fish is found in southern New England waters, and as the exact nature of the changes found in its pituitary gland is still to be established, we can draw no conclusions regarding the time or the regions of breeding activity of the swordfish at present. Future examinations of the reproductive condition of this fish in all portions of its range will undoubtedly aid in the solution of this problem.

I am deeply in debt to Dr. A. B. Dawson of the Harvard Biological Laboratory for encouragement and aid in pursuing this study; to the staff members of the Woods Hole Oceanographic Institute, for their generosity in providing laboratory space and equipment; and to Mr. Samuel Cahoon of Woods Hole, whose facilities and knowledge of swordfishing were invaluable in securing the material.

SUMMARY

Twenty-three swordfish pituitaries were studied in 1940. It was impossible to determine the sex of these fish; therefore the work was repeated in 1941 on pituitaries from 13 swordfish, all of which proved to be female. Their pituitaries were identical with those obtained in 1940.

The swordfish hypophysis is a blunt pyramidal structure, situated with its base in close approximation with the infundibulum. The pars nervosa descends from the infundibular floor, as a main or primary infiltration which arborizes in the ventral portion of the gland, with smaller secondary infiltrations of infundibular tissue penetrating the more dorsal regions. It is fibrous in nature, with the nuclei of connective tissue cells occurring frequently. Herring's bodies are not found in the pars nervosa, and there is no evidence of migration of cells from other glandular regions into it.

The pars intermedia surrounds the arborized part of the main nervous penetration, as a cone-shaped region with its apex directed ventrally. There are three types of cells in this part of the gland; an orange G acidophile, a weak blue basophile, and a chromophobe. Carmine granules are found in many of the basophiles. The acidophiles

become more numerous in glands obtained successively during the summer.

The Übergangsteil is a wedge-shaped region, lying dorsal to the base of the cone-like pars intermedia, with its main portion in the postero-dorsal part of the hypophysis. A sheath of its material completely envelops the pars intermedia. The Übergangsteil possesses two types of cells: a strong carminophile with finely granulated cytoplasm, and a deep blue basophile with coarsely granulated cytoplasm. During the latter half of the summer the basophiles contain large vacuoles which first appear about the middle of July.

The pars anterior occupies a horse-shoe shaped region in the antero-dorsal part of the hypophysis. Its arms extend posteriorly toward the main mass of the Übergangsteil, often surrounding the zone of contact of the infundibulum and the pars glandularis in collar-like fashion. The pars anterior of the swordfish pituitary has two chief types of cells: an amphiphile and an acidophile. The former are of a bluish-purple color, and are found bordering the secondary infiltrations of infundibular material that enter this region. The acidophiles are coarsely granulated carminophiles, which lie beneath the more peripheral amphiphiles to make up the bulk of the pars anterior.

The 13 swordfish from which gonads were obtained in 1941 were females. The ovaries contained ova which did not change appreciably in diameter during the summer. Thus there is no evidence of sexual maturation of swordfish taken in southern New England waters.

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